CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA

PEBULATE

Chemical Code # 000590, Tolerance # 00238
SB 950 # 345

April 6, 1987
Revised 2/2/88; 5/4/88; 4/20/90; 5/24/91; 5/31/96; 1/21/98

I. DATA GAP STATUS

Combined rat (onco & chronic): No data gap, possible adverse effect

Chronic dog: No data gap, possible adverse effect

Onco mouse: No data gap, no adverse effect

Repro rat: No data gap, no adverse effect

Teratogenesis rat: No data gap, no adverse effect

Teratogenesis rabbit: No data gap, no adverse effect

Gene mutation: No data gap, no adverse effect

Chromosome aberration: No data gap, no adverse effect

DNA damage: No data gap, no adverse effect

Neurotoxicity: Not required at this time[1]

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[1] - A possible adverse effect was noted in an acceptable study in the rat for necrosis in the brain. No comparable effect was found in an acceptable subchronic study in the rat.
Toxicology one-liners are attached.

All volume/record numbers through 052/158216 were examined.
** indicates an acceptable study.
Bold face indicates a possible adverse effect.

File name: T980121

Index prepared by R. Marovich and summary by B. Davis 4/6/87.
Summary revised by J. Gee, 5/3/88; J. Kishiyama & M. Silva, 4/20/90; Kishiyama & Silva, 5/24/91; H. Green & M. Silva, 5/31/96; Silva, 1/21/98.

II. SUMMARY OF TOXICOLOGY DATA

COMBINED CHRONIC/ONCOGENICITY, RAT

**** 025 074658**, "A Two Year Chronic Toxicity/Oncogenicity Study of Tillam Technical in Rats," (Daly, I.W., Bio/dynamics, Inc., Laboratory Report No. T-12733, 5/11/89). Tillam® technical (purity = 97.3%; LOT #: WRC 4921-20-18) was administered in the feed at concentrations of 0, 15, 150 or 1500 ppm to 70 CD® (Sprague-Dawley derived) rats/sex/group for 2 years. **Possible adverse effect**: An increased incidence of zonal dysjunction in the lens, retinal degeneration and posterior subcapsular cataracts was observed at 1500 ppm in both sexes. In males at 1500 ppm, there was an increased incidence of hemorrhage in thymus, spinal cord and liver, necrosis in liver and pulmonary edema--noted primarily in unscheduled deaths. Mortality was significantly increased at 1500 ppm in males. Systemic NOEL = 15 ppm/day (Eye effects--see above; hemoglobin in females and hematocrit in both sexes were decreased at 1500 ppm; body weight was less (11.4% - 39.8%) than in controls at $\geq$ 150 ppm from the first month of the study until termination). Oncogenic NOEL $\geq$ 1500 ppm/day. ACCEPTABLE. (Kishiyama & Silva, 4/12/90).

CHRONIC TOXICITY, **RAT**

004 024189, "56 Week Feeding Study in Rats". (Hazleton labs, 11/10/78); Tillam® technical, lot# 3905-36, 98.2% stated purity; 60 rats/sex/group at 0, 5, 20, or 80 mg/kg diet; at end of week 12 dose increased for 10 females from 80 to 160 mg/kg, in week 16 twenty rats/sex in the 80 mg/kg group and 25 rats/sex in the 20 mg/kg group placed on control diet; no adverse effects indicated. Incomplete, UNACCEPTABLE--excessive early mortality (12/60 high dose males died of hemorrhage during weeks 9-14 of study), altered blood clotting parameters, multiple dose changes, less than two years . (Apostolou, 8/30/85).

CHRONIC TOXICITY, DOG

** **024 072406,** "One-Year Toxicity Study with Tillam* in Beagle Dogs", (Pettersen, J.C. & Taylor, D.O.N., I.C.I. Americas Inc., Environmental Health Center, Report no. T 13000, 12/9/88). Pebulate technical (purity = 97.3%; WRC #: 4921-20-18; I.D. #: 0751-33) was administered orally (capsules) at concentrations of 5, 25, 50 or 100 mg/kg/day to 4 Beagle dogs/sex/group for 1 year. NOEL = 5 mg/kg/day (significant increase in hemolysis of red blood cells in both sexes at $\geq$ 25 mg/kg/day (females) or $\geq$ 50 mg/kg/day (males) with associated increase in the severity of hemosiderin deposition in the liver and spleen; pathological effects on the spinal cord in the nervous system were also observed). **Possible adverse effects**: hemolysis of the red blood cells and possible degenerative effects (Wallerian-type degeneration) were observed in spinal cord (both sexes). ACCEPTABLE. (Kishiyama & Silva, 4/11/90).

022 069212 This volume contains a letter regarding spinal cord lesions observed in a preliminary update of the chronic dog study 072406. No work sheet. (Kishiyama & Silva, 4/10/90).

ONCOGENICITY, **MOUSE**

Subchronic Studies:

030 096621, "Pebulate: 28 Day Feeding Study in Mice", (D.J. Tinston, ICI Central Toxicology Laboratory, CTL Study No: PM0723, Report No: CTL/T/2644, 5/15/89). Pebulate technical (purity = 97.1%) was administered in the feed at concentrations of 0 (diet), 300, 1000, 2500, or 7000 ppm to 10 mice (C57BL/10JfCD-1/Alpk)/sex/group for 28 days. The 7000 ppm group was terminated due to excessive loss of bodyweight thought to be due to unpalatable food. NOEL = 300 ppm (Decreased bodyweight gain for both sexes at 2500 ppm (approximately 10%). Food consumption was reduced approximately 10% for both sexes at 2500 ppm group. Hemoglobin, hematocrit, mean cell hemoglobin for both sexes at 2500 ppm and mean cell volume for both sexes at $\geq$ 1000 ppm. Adjusted liver weight in males at 1000 ppm and in both sexes at 2500 ppm were increased. The incidence of accentuated lobular pattern of the liver in males and reduced corpora lutea in females at 2500 ppm group was considered treatment related. (Kishiyama & Silva, 5/15/91).

Oncogenicity Studies:

** 031 096622, "PEBULATE: 18 Month Carcinogenicity Study in Mice", (D. J. Tinston, ICI Central Toxicology Laboratory, Report No: CTL/P/3119, Study No: PMO729, 3/14/91). Pebulate technical (purity = 97.1%) was administered in the feed at concentrations of 0 (diet), 300, 1000, or 3000 ppm to 50 C57BL/10JfCD-1/Alpk mice/sex/group for 18 months. NOEL = 300 ppm/day (Reduced body weight gain and food consumption was observed in both sexes at 3000 ppm. Palatability may have affected diet consumption at 3000 ppm. Increased platelet counts were reported in both sexes at $\geq$ 1000 ppm--29% and 34% at 3000 ppm for males and females, respectively. Increased white blood cell (32%) and lymphocyte counts (33%) were observed in males at 3000 ppm. Increased liver weight and incidence of hepatocyte fat vacuolation was observed in both sexes at $\geq$ 1000 ppm.) No oncogenic effects were observed at any dose. ACCEPTABLE. (Kishiyama & Silva, 5/16/91).

004, 006, 012, 015 024187, 037367, 055171, 059846, "Lifetime Oral Study in Mice"; (IRDC, 4/17/80). Tillam, technical, 98.2%, lot 3905-36; fed in the diet to 60/sex/group at 0, 10, 40 and 160 mg/kg/day for 2 years; 10/sex/group for interim sacrifice; NOEL $\geq$160 mg/kg/day; no adverse effect reported, no oncogenic effect; #037367 contains individual data, #055171 contains diet analysis, #059846 contains group food consumption; UNACCEPTABLE (inadequate high dose). Apostolou, 8/29/856; Davis, 7/29/86, 3/31/87; Gee, 5/3/88).

REPRODUCTION, **RAT**

** 023, 032 072842, 092460 "Two Generation Reproduction Fertility Study in Rats", (Keller, K.A., I.C.I. Americas Inc., laboratory project ID 152-96, January 19, 1989). Pebulate (purity 97.3%; WRC 4921-20-18) was administered in the feed at concentrations of 0, 15, 120 or 1000 ppm and fed to 26 Charles River COBS® CD® rats/sex/group during pre-mating through lactation. Parental NOEL = 15 ppm (High mortality for F0 males and both sexes of F1 at 1000 ppm; hemorrhage was observed subcutaneously and in several organs in F0 males and both sexes of F1 at 1000 ppm; reduced food intake was observed in males of F0 and F1 at $\geq$ 120 ppm and in females at 1000 ppm; erythrocyte count, hemoglobin concentration, % hematocrit and mean corpuscular hemoglobin concentration were decreased while platelet count was increased in F0 females at $\geq$ 120 ppm and F1 females at 1000 ppm

(too few F1 males were available to assess at 1000 ppm); increased platelet count was observed). Developmental NOEL = 120 ppm (Reduced body weight and increased mortality were observed in F1a and F1b pups at 120 ppm. No F2a or F2b pups were produced at 1000 ppm). Originally reviewed as unacceptable (Silva, 4/17/90), upon submission of the requested information (dose justification, analyses of pebulate concentrations in the diet, Table 44, explanation of pup viability data (page 74) and a clarification between the statement on page 30 (# of pups found dead during lactation) and data in Table 20), the study is now **acceptable.** M. Silva, 5/20/91.

TERATOGENICITY, **MOUSE**

004 024186, "Tillam-Safety Evaluation by Teratological Study in the Mouse". (Woodard Research Corporation, 4/28/67); Tillam technical, lot# 2698205 (96.2% purity); 0, 8 or 24 mg/kg; No adverse effects. Maternal NOEL $\geq$ 24 mg/kg (HDT); Developmental NOEL $\geq$ 24 mg/kg (HDT). Incomplete, UNACCEPTABLE--only two doses without justification of dose levels, no evidence of toxicity at the high dose, no information on the strain or age of animals, no information on the analysis of the test material or the dosing solution, no necropsy data, no individual skeletal anomaly data, no quality assurance, only summary data on fetal anomalies. Original review (Apostolou, 8/29/85) found insufficient data to discount possible adverse effects. Rereview with additional data (Davis 7/29/86) found no adverse effect.

004 024185. Addendum to 024186.

007 037368. Supplement to 024186.

TERATOLOGY, RAT

**010 051395, "A Teratology study in rats with Tillam Technical" 833. (WIL Research Labs, Study # 27034, 11/12/86) Pebulate Technical WRC #4921-20-18, purity = 97.3%; 0, 5, 30, or 200 mg/kg/day on days 6-15 by oral gavage to 25 animals each group; reduced ossification of sternebrae and 13th rib in high dose fetuses associated with decreased maternal weight gains; reduced fetal weight in high dose group. Maternal NOEL (reduced weight gain) = developmental NOEL (Degree of ossification and reduced fetal weight) = 30 mg/kg/day. No adverse effect, complete, ACCEPTABLE. (Davis 3/30/87).

TERATOLOGY, RABBIT

**010 051394, "A Teratology study in Rabbits with Tillam Technical;" 833; (WIL Research Labs, Study # 27036, 11/24/86) Pebulate Technical WRC #4921-20-18, 97.3% purity; 0, 5, 30, or 150 mg/kg/day in 0.5 ml/kg on days 7-19 by oral gavage to 20 animals each group; high dose caused maternal weight loss; no fetotoxic, embryotoxic or developmental affects observed. Maternal NOEL = 30 mg/kg/day (weight loss); Developmental NOEL $\geq$ 150 mg/kg/day (HDT). No adverse effect, complete, ACCEPTABLE. (Davis 3/27/87).

GENE MUTATION

007 037369, "Mutagenicity Evaluation of Tillam Tech CGC-0201--Final Report. Gene Mutation, Ames (842); DNA Damage, Yeast Gene Conversion (844); (Litton Bionetics, 10/77); Tillam technical lot# CGC-0201 (no purity stated); 0.001, 0.010, 0.100, 1.000, or 5.000 ul; No adverse effect. Incomplete, UNACCEPTABLE, no information on test material, no plate replicates, no confirming experiment, no quality assurance information, solvent used for test material and negative controls ambiguous. Even less information is presented for the yeast gene conversion assay: no experimental design, no evaluation criteria, no evaluation of the adequacy of the positive control with activation. No adverse effect indicated. (Davis 7/25/86).

** 013 058588, "Mutagenicity Evaluation in Salmonella typhimurium - Tillam", (Stauffer Chemical, CT, 6/19/87). Salmonella typhimurium, tested with pebulate, 97.3%, lot 4921-20-18, strains TA1535, TA1537, TA98 and TA100, at 0, 0.0375, 0.075, 0.15, 0.30 and 0.60 ul/plate, triplicate plates, two trials with each strain; tested to the limit of solubility with some cytotoxicity; with and without Aroclor-induced rat liver activation; no evidence of increase in reversion rate; ACCEPTABLE. (Gee, 5/3/88).

CHROMOSOME ABERRATIONS

** 028 086234, "Pebulate: An Evaluation in The In Vitro Cytogenetic Assay in Human Lymphocytes," (Randall, V. & Mackay, J.M., ICI Central Toxicology Laboratory, Report No. CTL/p/2826, 1/9/90). Pebulate technical (purity = 97.1%; Ref #: WRC 11321-14 & 4921-20-18) was used at concentrations of 0 (DMSO), 15, 75, and 150 ug/ml with and without S-9 (from Aroclor 1254-induced male Alpk:APfSD rats) on human male and female lymphocytes for 3 hours. Note: female lymphocytes without S-9 were treated at 100 (not 150) ug/ml. Pebulate did not statistically increase the number of chromosomal aberrations or produce a positive response. ACCEPTABLE. (Kishiyama & Silva, 4/10/90).

013 058589, "Mutagenicity Evaluation in L5178Y Mouse Lymphoma Multiple Endpoint Test: Cytogenetic Assay ", (Stauffer, CT, 6/18/87 T-12889). Chromosomal aberration assay with Tillam, 97.3%, amber liquid; tested without activation at 0, 0.03, 0.04, 0.05, 0.06 and 0.07 ul/ml, in duplicate cultures; tested with rat liver activation at 0.006, 0.008, 0.01, 0.012 or 0.014 ul/ml, 4 hours followed by 8 additional hours; single harvest time; scored 50 cells per culture, 100 per concentration; no evidence for increase in aberrations; gaps scored but not reported; positive control (DMN) for activation showed marginal response; UNACCEPTABLE (poor response with positive control for activation). Not upgradeable. (Gee 5/3/88).

DNA DAMAGE/OTHER

** 029 088827, "Pebulate: Assessment for the Induction of Unscheduled DNA Synthesis in Rat Hepatocytes In Vivo", (J. C. Kennelly, ICI Central Toxicology Laboratory, Report No. CTL/P/3032, Study No. SR0416, 8/17/90). Pebulate technical (purity = 97.1%; Ref #: WRC 11321-14 & 4921-20-18) was administered as single oral dose (gavage) at concentrations of 200, 400, or 800 mg/kg to male Alderley Park (Alpk:APfSD) rats with sacrifice times at 4 or 12 hours (5 rats/dose/time point--40% or 60% of rats/group were sacrificed 4 hours and the rest at 12 hours after dosing). Negative corn oil control and postive 6-p-dimethylaminophenylazobenzthiazole, or N-nitrosodimethylamine controls had 4 treated rats/group but only one sacrificed/time point (4 and 12 hours). 100 cells/rat/dose/time point were examined for UDS. The test was performed twice. There was no increase in UDS at any dose of pebulate. **Acceptable.** (Kishiyama & Silva, 5/15/91).

** 013 058589, "Mutagenicity Evaluation in L5178Y Mouse Lymphoma Multiple Endpoint Test: Cytogenetic Assay (SCE)", (Stauffer, 6/18/87 T-12889); Tillam, 97.3%, lot no. 4921-20-18; L5178Y tested for sister chromatid exchange without activation at 0, 0.03, 0.04, 0.05, or 0.06 ul/ml and with rat liver activation at 0.004, 0.006, 0.008, 0.10 or 0.12 ul/ml, 4 hours treatment followed by 22 hours with BrdU with colcemid for 2 hours; scored 50 cells in each culture for a total of 100 cells peer concentration; no evidence of an increase in sister chromatid exchanges with or without activation; ACCEPTABLE. (Gee, 5/3/88).

NEUROTOXICITY

004 024188; 005 037366, "Acute Delayed Neurotoxicity Study with Technical Tillam in Adult Hens." 817 (Stauffer Richmond Toxicology Laboratory, 10/16/80); Tillam technical, lot #CGB-0201 (95% purity); 0 or 8873 mg/kg; No adverse effect indicated. Neurotoxic NOEL > 8873 mg/kg (HDT). Incomplete, UNACCEPTABLE--Negative control animals have an array of abnormalities which confounds the entire study. 037366 is the same as 24188 but with additional data. (Apostolou, 8/30/85; Davis, 7/28/86).

**** 045, 050 & 051 130010, 158209 & 158215,** "Pebulate: Acute Neurotoxicity Study in Rats," (S.A. Horner, Zeneca Central Toxicology Laboratory, Cheshire, UK., Report # CTL/P/4181, 3/29/94). "First Revision to Pebulate: Acute Neurotoxicity Study in Rats," (Horner, S.A, Zeneca CTL, Cheshire, UK, 12/8/95), and "Thiocarbamates: Selective Re-examination of Neuropathology," (Chalmers, D.T., Duffell, S.J. and Horner, S.A., Zeneca CTL, Cheshire, UK, 3/28/95). Technical pebulate (96.1% (w/w) pure) was administered by gavage to Alpk:APfSD rats (10/sex/dose) in a single dose at 0 (corn oil), 50, 150, and 500 mg/kg followed by a 15-day observation period. Systemic NOEL = 50 mg/kg (There was an increase in clinical signs in both sexes at 500 mg/kg (decreased activity, irregular breathing, hunched posture, piloerection, ptosis, splayed gait, salivation, urinary incontinence, and decreased visual placement response). Group mean bodyweights at $\geq$ 150 mg/kg (males) were significantly decreased. Group mean food consumption was reduced at 500 mg/kg (males) during week 1 compared to controls.) Neurotoxicity NOEL = 150 mg/kg (Landing foot splay was increased at 500 mg/kg (females) on days 1, 8, and 15. Increased time to tail flick was noted at 500 mg/kg (both sexes) on day 1. Microscopy revealed neuronal cell necrosis in the pyriform cortex and in the dentate gyrus of the brain at 500 mg/kg (both sexes). **Possible adverse effect.** Initially reviewed as having no adverse effect (Silva, 5/28/96), in light of the slide re-examination (Record #: 158209) the study was flagged for a possible adverse effect. **Acceptable**. (Silva, 1/21/98).

** 046, 050 & 052 130305, 158209 & 158216 "Pebulate: Subchronic Neurotoxicity Study in Rats", (A. Brammer, Zeneca Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK, Report # CTL/P/4246, 21 April 1994). "First Supplement to: Pebulate: Subchronic Neurotoxicity Study in Rats," (Brammer, A., Zeneca CTL, Cheshire, UK, 11/30/95). "Thiocarbamates: Selective Re-examination of Neuropathology," (Chalmers, D.T., Duffell, S.J. and Horner, S.A., Zeneca CTL, Cheshire, UK, 3/28/95). Pebulate technical (96.1% w/w pure) was fed in diet to Alpk:APfSD rats (12/sex/dose) at 0, 50, 250, and 1000 ppm (w/w) in the diet for 13 weeks. Systemic/Neurotoxicity NOEL (Nominal) = 50 ppm (Significant decreases in bodyweight occurred in both sexes at $\geq$ 250 ppm. Food consumption was decreased in both sexes, primarily at 1000 ppm. At $\geq$ 250 ppm females showed statistically significant increases in landing foot splay week 5. Time to tail-flick was decreased (statistically significant) for females at 250 ppm week 5 and for females at $\geq$ 250 ppm at week 9. Group mean brain weight relative to bodyweight was reduced 5% in males at $\geq$ 250 ppm and 3% in females at 1000 ppm. Also, brain width and width adjusted for bodyweight were reduced in females at 1000 ppm. Sciatic nerve fiber degeneration

(minimal) was increased for both sexes at 1000 ppm.) ChE NOEL (Nominal) = 50 ppm (Brain cholinesterase activity was reduced (statistical significance) for high dose males (13%) and females (23%) and for mid-dose males (8%) compared to controls.) No adverse effect. Acceptable. (Silva, 1/21/98).

ADDITIONAL STUDIES:

047 135131 "Thiocarbamates: Comparative In Vivo Percutaneous Absorption Study in The Rat," (Lythgoe, R.E. and J.A. Platt, Zeneca Central Toxicology Laboratory, Cheshire, UK, Report No: CTL/P/4594; Study No: UR0445; 1/12/95). Pebulate, EPTC, cycloate, butylate and vernolate (each at 0.3mg, replicated 4 times,) were administered one time only as emulsifiable concentrates (1/100 aqueous dilution of a [14C]-labelled) to non-occluded skin of CD Crl:CD(SD)BR rats (10/compound). Evaluation was 10 hours after treating. Recovery of radioactivity averaged 76.1%, 93.7%, 94.9%, 95.4% and 92.9% for EPTC, cycloate, butylate, pebulate and vernolate, respectively. The high proportion of the applied dose for all 5 thiocarbamates was from the non-occlusive cover, indicating that a large percentage of the applied dose volatilized from the skin surface (captured by active carbon filters). EPTC recovery was comparatively lower due to its greater volatility and the volatility occurred during dosing, rather than during the 10 hour exposure. Cycloate absorption (material found in systemic circulation) was 15.90%, compared with 9.9%, 7.35%, 5.76% and 4.0% for pebulate, vernolate, EPTC and butylate, respectively. For pebulate, radioactivity was recovered in the following percentages: Skin wash, 4.26%; non-occlusive cover, 79.37%; application site, 1.72%; untreated skin, 0.09%; total not absorbed, 85.45%; urine, 3.11%; feces, 0.14%; cage wash, 0.40%; carbon dioxide, 1.32%; expired volatiles, 0.14%; carcass, 4.81%; total absorbed dose, 9.90%; total recovered, 95.35%. (Kishiyama & Silva, 5/31/96).